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Comparing adult hippocampal neurogenesis in mammalian species and orders: influence of chronological age and life history stage

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Adult hippocampal neurogenesis is a prominent event in rodents. In species with longer life expectancies, newly born cells in the adult dentate gyrus of the hippocampal formation are less abundant or can be completely absent. Several lines of evidence indicate that the regulatory mechanisms of adult neurogenesis differ between short- and long-lived mammals. After a critical appraisal of the factors and problems associated with comparing different species, we provide a quantitative comparison derived from seven laboratory strains of mice (BALB, C57BL/6, CD1, outbred) and rats (F344, Sprague-Dawley, Wistar), six other rodent species of which four are wild-derived (wood mouse, vole, spiny mouse and guinea pig), three non-human primate species (marmoset and two macaque species) and one carnivore (red fox). Normalizing the number of proliferating cells to total granule cell number, we observe an overall exponential decline in proliferation that is chronologically equal between species and orders and independent of early developmental processes and life span. Long- and short-lived mammals differ with regard to major life history stages; at the time points of weaning, age at first reproduction and average life expectancy, long-lived primates and foxes have significantly fewer proliferating cells than rodents. Although the database for neuronal differentiation is limited, we find indications that the extent of neuronal differentiation is subject to species-specific selective adaptations. We conclude that absolute age is the critical factor regulating cell genesis in the adult hippocampus of mammals. Ontogenetic and ecological factors primarily influence the regulation of neuronal differentiation rather than the rate of cell proliferation.

Introduction

The de novo formation and integration of new neurons into the existing cortical circuitry is an extreme along the spectrum of plastic changes that permit the adult mammalian brain to adapt to changing functional demands (reviewed in Leuner & Gould, 2010). Adult hippocampal neurogenesis results in the formation of new granule cells in the dentate gyrus that extend dendrites (reviewed by Ribak & Shapiro, 2007) and axons to the usual targets of this cell type (Stanfield & Trice, 1988; van Praag et al., 2002). The cells become electrophysiologically integrated (van Praag et al., 2002) and their immediate early gene reaction to internal and external stimuli is similar to that of resident granule cells (Okano et al., 1993; Ramirez-Amaya et al., 2006; Kee et al., 2007; Snyder et al., 2009). Not all newly formed cells survive; many of them die within weeks after generation (Dayer et al., 2003). The formation and functional integration of a large number of cells, many of which die shortly after, has led to the idea that 'young neurons' may be transient, functionally distinct elements of the hippocampal circuitry (Cameron & McKay, 2001; Amrein et al., 2004b). This idea is supported by the changing electrophysiological and morphological properties of the neurons from the young to the adult stage (reviewed in Mongiat & Schinder, 2011). During maturation, the young neurons recapitulate the developmental stages in the responsiveness to GABA (Ambrogini et al., 2004; Espo´sito et al., 2005). Once in the neuronal lineage, young neurons are less inhibited by GABA than mature granule cells (Wang et al., 2000). The higher excitability of young granule cells, which still have short apical and basal dendrites, leads to robust long-term potentiation at a lower induction threshold than mature granule cells (Schmidt-Hieber et al., 2004). Young neurons are also preferentially recruited upon (behavioural) activation of the hippocampus, possibly due to the increased release of GABA resulting in the activation of type 2 progenitor cells (Tozuka et al., 2005; Ramirez-Amaya et al., 2006; Kee et al., 2007; Snyder et al., 2009). In summary, new granule cells are more excitable than mature, resident granule cells, and cells at all stages of neurogenesis (proliferation, neuronal differentiation and survival) are subject to a

plethora of physiological, neurochemical, hormonal and behavioural regulatory influences (reviewed by Abrous et al., 2005).
A marked decline in adult hippocampal neurogenesis with ageing has been described in mice, rats and primates including humans (Kuhn et al., 1996; Cameron & McKay, 1999; Gould et al., 1999; Leuner et al., 2007; Knoth et al., 2010; Ben Abdallah et al., 2010). So far, it has not been assessed if the decline in neurogenesis can be compared in animals that differ markedly in life span and that reach equivalent life history stages at very different absolute ages. Here, we investigate the effect of differences in development and life history on hippocampal neurogenesis by collecting and analysing data of cell proliferation and neuronal differentiation in short- and long-lived mammals. With this meta-analysis, we aim to clarify whether the extent of adult hippocampal neurogenesis follows a general or species-specific pattern with chronological age, and whether the regulation of neurogenesis is linked to life history events.

A comparative survey

Adult hippocampal neurogenesis has now been confirmed in freeliving rodents (Galea & McEwen, 1999; Lavenex et al., 2000; Amrein et al., 2004b; Epp et al., 2009) and other mammals (summarized in Amrein et al., 2008). Adult hippocampal neurogenesis has also been found in insectivores (Bartkowska et al., 2008; Alpa´r et al., 2010; Bartkowska et al., 2010). However, in shrews, adult hippocampal neurogenesis is literally switched off in the first winter experienced by the animals and remains off for the remainder of the animal's life (Bartkowska et al., 2008). Taxonomic groups that are longer lived than rodents or shrews exhibit, in general, low levels of adult hippocampal neurogenesis. Bats are peculiar as adult hippocampal neurogenesis was found to be absent in a large species sample of Microchiroptera (Amrein et al., 2007). Megachiroptera (flying foxes or fruit bats) do show adult hippocampal neurogenesis, but at a very low level (Gatome et al., 2010).
Similar observations have been made in primates. Adult hippocampal neurogenesis is observed in both New World monkeys (Gould et al., 1998; Leuner et al., 2007) and Old World primates (Gould et al., 1999; Kornack & Rakic, 1999; Perera et al., 2007) including humans (Eriksson et al., 1998). In contrast to rodents, the number of newly generated neurons is very small after the onset of puberty in both non-human primates (Jabe`s et al., 2010) and humans (Knoth et al., 2010). It is striking in view of the very low proliferation that there are more young neurons in primates than would be expected from the results of rodent studies (Taffe et al., 2010). Foxes are similar to primates in that they show low proliferation and prolonged expression of markers for young neurons (Amrein & Slomianka, 2010), whereas cell proliferation in dogs results in the formation of few young neurons in young animals and proportionally even fewer in aged animals (Siwak-Tapp et al., 2007). Both carnivores differ from laboratory rodents, in which the proliferation and neuronal differentiation decline with age but maintain a rather constant relation (Ben Abdallah et al., 2010).
So far, the data indicate that (i) some mammalian species show normal behaviour without adult hippocampal neurogenesis, (ii) adult hippocampal neurogenesis is ontogenetically regulated in a speciesspecific manner, (iii) species-specific regulatory mechanisms control transitions between the cellular stages of cell proliferation, survival and neuronal differentiation, and (iv) adult hippocampal neurogenesis of long-lived species is characterized by low proliferation rates. In this review, we address how the ontogenetic regulation of adult hippocampal neurogenesis differs between species and orders, and formulate tentative conclusions and prospective ideas from these observations. As a starting point, we will outline general set points of granule cell genesis and ontogenetic landmarks.

General time-dependent features of granule cell genesis in the dentate gyrus of mammals

Early development

In the hippocampal formation, the densely packed granule cells form the principal cell layer within the dentate gyrus (Amaral et al., 2007). Within the hippocampal formation, the dentate gyrus is the last structure to be fully formed (Bayer, 1980b). In rats, the first granule cells appear at embryonic days 14–20, depending on the strain (Schlessinger et al., 1975; Bayer, 1980b). Between 15 and 20% of all granule cells are generated prenatally, and the remaining large majority of granule cells are formed between birth and postnatal day 18. Only 5–10% are generated afterwards (Bayer, 1980a). Similar time courses have been described in mice, where granule cell generation starts at embryonic day 10, peaks at birth and tapers off at postnatal day 20 (Angevine, 1965). In precocial spiny mice that are born after a prolonged gestation period (38 day), the morphology of the dentate gyrus is already mature-like at birth and slight volumetric growth can be observed only until postnatal day 10 (Brunjes, 1984). In the guinea pig, which is also precocial but has an even longer gestation (67 day) than spiny mice, neurogenesis is still prominent after birth but accounts for only _20% of the total granule cell number, and declines sharply after 3 weeks (Guidi et al., 2005). In primates, similar to precocial rodents, around 85% of the granule cells are generated prenatally (summarized by Seress, 1992). The same author estimates that proliferating cells in the human dentate gyrus amount to around 0.4% relative to the total granule cell population at the time of birth, and proliferation decreases steadily afterwards (Seress et al., 2001). Whether proliferation of granule cells peaks pre- or postnatally seems to depend on the developmental state of a given species at birth. A mostly prenatal development of the dentate gyrus granule cells facilitates the rapid maturation of the hippocampus after birth, as required in precocial rodents such as spiny mice and guinea pigs. Primates including humans also develop most of their granule cells prenatally, but it is not known when the hippocampus achieves its full functional competence.

From developmental to adult neurogenesis

It is difficult to draw a strict border between what should be considered as neurogenesis during ontogenetic development and what should be termed adult neurogenesis. The two processes are probably overlapping, and consistent with this gradual shift there is also a debate about when an animal should be considered adult (Spear, 2004) (for important considerations of ageing and neurogenesis across vertebrates see Lindsey & Tropepe, 2006). We and others have shown that neurogenesis in rodents declines exponentially with age shortly after birth (Kuhn et al., 1996; Cameron & McKay, 1999; Rao et al., 2006; Ben Abdallah et al., 2010). In contrast, the amount of dying cells follows a probability density function that peaks at just prior to 2 months of age in mice (Ben Abdallah et al., 2010). The ratio between dying and proliferating cells at 1 month of age is very low, indicating that at this time many of the newly generated cells survive (Ben Abdallah et al., 2010). This observation was taken as an indication that neurogenesis up to 2 months marks the tail-end of postnatal neurogenesis in mice. A possible shift between postnatal and adult neurogenesis between 1 and 2 months of age is supported by changes in the genetic composition of intrinsic factors within the precursor cell population from a postnatal to an adult phenotype, which might take until postnatal day 60 in mice to be fully established (Gilley et al., 2011). A fast decline of neurogenesis with age is also documented for primates (Gould et al., 1999; Leuner et al., 2007; Jabe`s et al., 2010), but the time course of down-regulation has not been assessed from a comparative point of view.

Neurogenesis in aged animals

In middle-aged to senescent rodents, neurogenesis is reduced to low levels and seems to be rather stable between 12- and 24-month-old rats (Heine et al., 2004a; Rao et al., 2005, 2006) and mice (Kronenberg et al., 2006). There is still a debate about whether the reduced neurogenesis is due to a decrease in the number of precursors (Olariu et al., 2007) or increased quiescence of the precursor cells (Hattiangady & Shetty, 2008). Cell death also decreases in aged animals, and the higher survival rate of proliferating cells in aged animals may be an adaptive response to low proliferation in laboratory mice (Ben Abdallah et al., 2010). The dependence of aged animals on a few newly generated neurons may also increase the survival rate in wild rodents (Amrein et al., 2004a). The morphological properties of the newly born cells in aged rodents are still similar to young rodents (van Praag et al., 2005; Morgenstern et al., 2008), but altered in aged primates (Aizawa et al., 2011). For an in-depth review of factors regulating neurogenesis in aged animals see Drapeau & Abrous (2008).

Ontogenetic landmarks

Adolescence (the transition between childhood and adulthood) is characterized by high impulsivity, high plasticity and the development of complex behavioural repertoires, ultimately leading to stable behavioural patterns that guarantee successful survival (for excellent review see Spear, 2000). Hallmarks of adolescence are increased social interactions with peers and risk- and novelty-seeking behaviour. In humans, Spear (2000) attributes this transition phase to the age span of 12–18 years. The duration of the adolescent phase in primates varies between species, but is well defined and displays many of the characteristic traits described in humans (Pereira & Altman, 1985; Steinberg et al., 1989). An adolescent (juvenile or subadult) phase is commonly assumed to last from postnatal day 28 to 42 in rats as demonstrated by behavioural, morphological and biochemical factors (Spear, 2000). Some researchers consider rats to be adolescent until postnatal day 60, when they normally reproduce for the first time (Maeda et al., 2000). In mice, the period of adolescence is not well defined as sexual maturity may be reached very soon after weaning. Reaching adult body size and the first successful breeding are two criteria marking the transition from adolescence to adulthood (Ernest, 2003). Concomitantly, the now adult animal has established its territory, providing the supplies pertinent for its own survival and that of its progeny. The full species-specific behavioural repertoires have been acquired, and changes thereof are unlikely, except when driven by external forces. However, having adult body size and reaching sexual maturity are not equivalent to having offspring. Depending on, e.g. social structure, environmental conditions, seasonality or animal husbandry, the time point of having the first offspring might differ considerably from the time when the animal reaches sexual maturity. Subordinate individuals of cooperative breeding species such as marmosets, foxes, meerkats and wolves may not reproduce for years after becoming sexually mature (Macdonald, 1979; Digby, 1995; Burkart et al., 2009). Even in fecund species with low reproductive skew, such as communally breeding house mice, elevated stress levels in breeding females delay the reproductive rates compared with females breeding in non-aggressive, preferred partner dyads (Weidt et al., 2008). In contrast to a delay in reproduction, in many species females start to reproduce before they achieve their adult body weight. Female common voles (Microtus arvalis) probably hold the record for the earliest reproduction in mammals; if environmental factors are favourable, they are capable of fertile mating by the age of 14 days (Tkadlec & Zejda, 1995). In meadow voles (Microtus pennsylvanicus), 30% of females are pregnant by the age of 30 days (Hamilton, 1937). Laboratory mice can be fertile as early as 28 days, but considerable strain differences have been reported (Berry & Bronson, 1992). Low ambient temperature most dictates mortality in wild mice, followed by death by predation (Berry & Bronson, 1992). Food

availability and ambient temperature, generally tightly interacting factors, determine offspring survival, reproductive output and adult survival. Aged rodents also show an increased vulnerability to low temperature due to increased homeostatic imbalance (Reynolds et al., 1985), and are more prone to stress- and age-related diseases (McEwen, 2002). Behavioural flexibility and activity decrease. The survival of the aged animal may depend, in addition to environmental conditions, on the social structure of the species, which can be supportive, indifferent or expelling.

A large-scale comparative approach

In order to investigate the effect of the differences in development and life history, we collected data on hippocampal neurogenesis in shortand long-lived mammals, and tested whether we could identify general or species-specific patterns in the regulation of cell proliferation and neuronal differentiation with chronological age and life history events. Before we discuss the findings, we would like to address the methodological limitations to the accuracy of the data.

Methodological considerations

A recurring impediment to evaluate the relation between neurogenesis, ecology, evolution and age is the reporting of data, which may have little functional significance in a comparative sense. Many articles report cell numbers as density measurements, which are able to show the effects of experimental conditions within the studied species or strains. Density measurements are, however, of little use in crossspecies comparisons due to varying methodology and protocols.
Furthermore, a short glimpse at the distribution and packing density of granule cells in rodents, foxes and primates (Fig. 1) illustrates that, within the same volume, different species can harbour varying numbers of granule cells. To us, the number of proliferating cells or young neurons relative to the resident granule cells, i.e. normalized cell numbers, signifies the relative importance of adult hippocampal neurogenesis, and it is the parameter that is used in the comparison made in the following (Figs 2 and 3). In studies where the total granule cell number was not reported together with neurogenesisrelated cell numbers, we supplemented it from other sources, assuming that total granule cell number is relatively stable with age in rodents and primates (Rapp & Gallagher, 1996; Calhoun et al., 1998; Keuker et al., 2003).
Another obstacle in comparative studies is differences in methodology for visualizing and assessing the numbers of neurogenesisrelated cells. In the comparative analysis presented here, we have only considered data that have been collected using design-based counting procedures spanning the whole septo-temporal extent of the dentate gyrus and evaluating the subgranular layer and granule cell layer. In addition, we have focused mainly on cell numbers derived from counts of cells expressing endogenous markers of proliferating cells

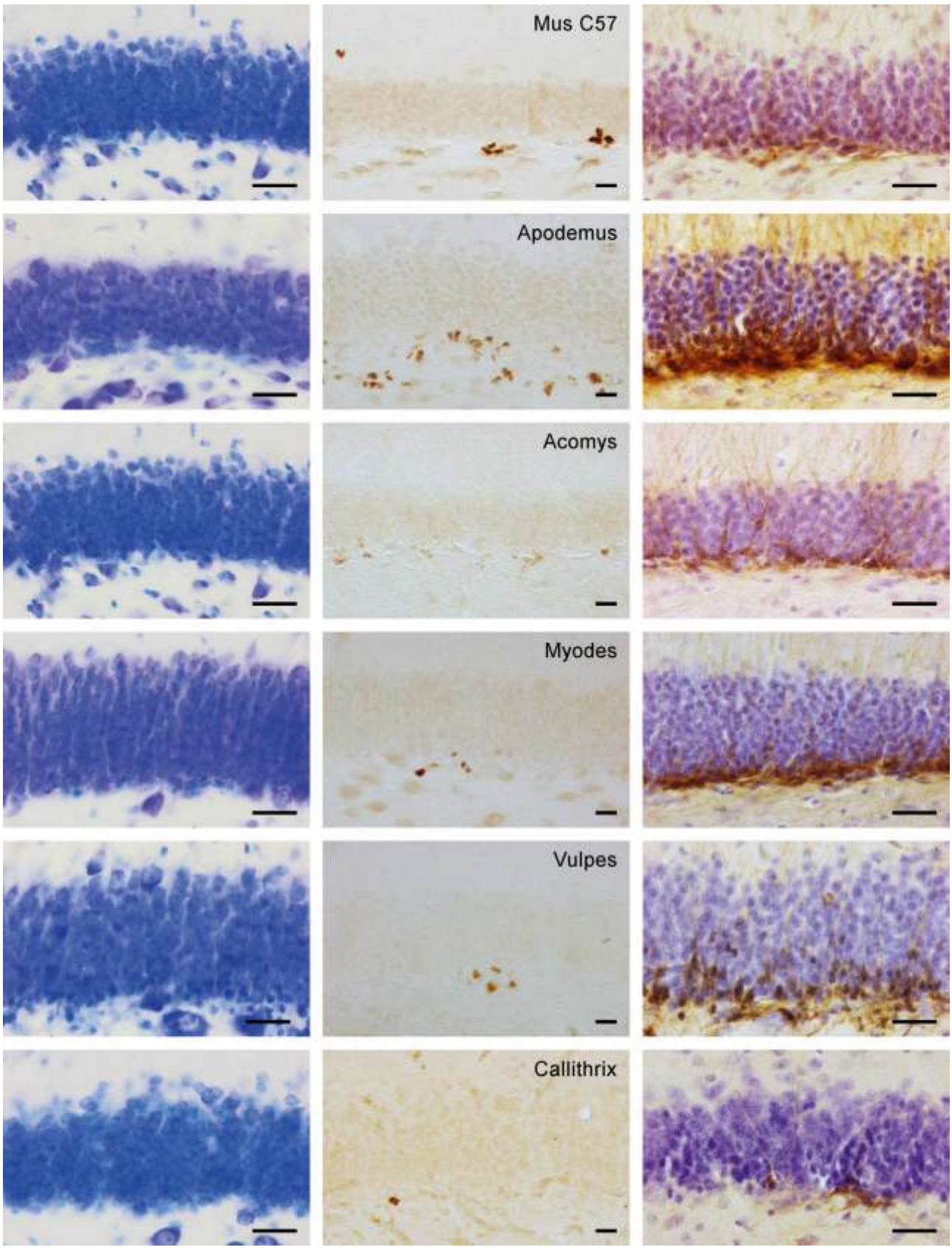
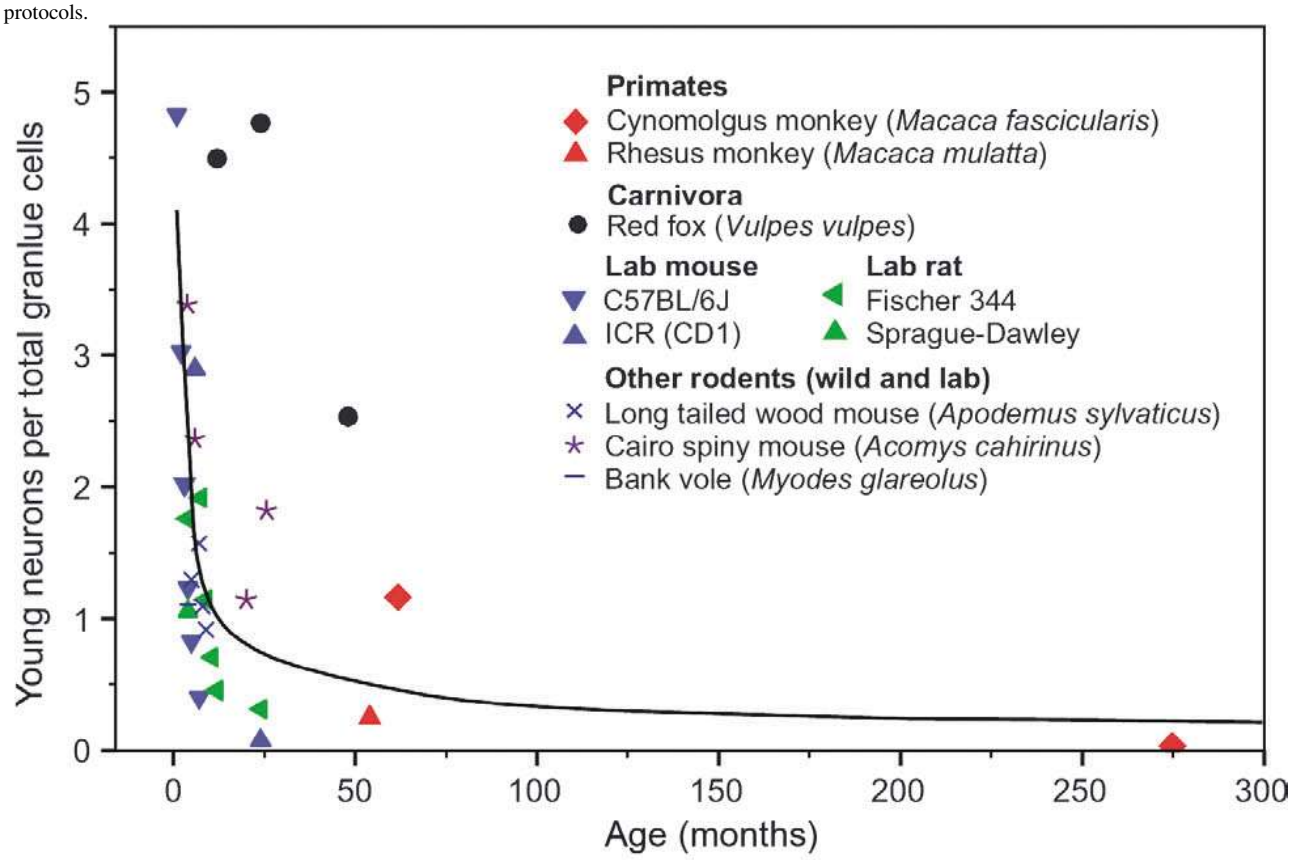
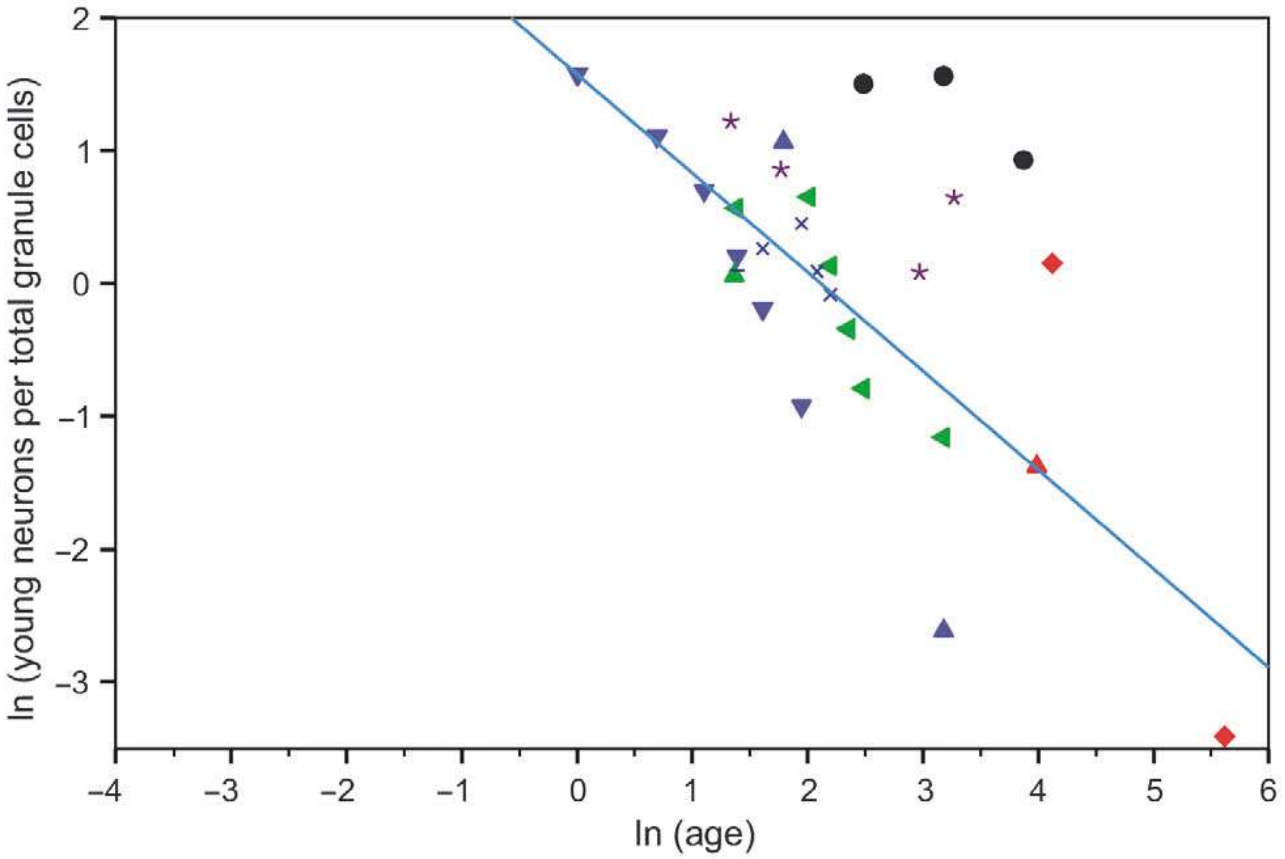


Fig. 1. Dentate gyrus morphology and neurogenesis across species. Methacrylate-embedded sections stained with Giemsa and immunohistochemically stained sections reveal differences in cell number, size and packing density of granule cells in the dentate gyrus in C57BL/6 (A–C), yellow-necked wood mouse (Apodemus flavicollis, D–F), spiny mouse (Acomys cahirinus, G–I), bank vole (Myodes glareolus, J–L), red fox (Vulpes vulpes, m–o) and common marmoset (Callithrix jacchus, P–R). Immunohistochemistry against Ki67, a chromosome-associated protein expressed during the active phase of the cell cycle (late G1–M) (Starborg et al., 1996; Scholzen & Gerdes, 2000; Kee et al., 2002), visualizes differences in the number and appearance of proliferating cells in the subgranular layer of the species (B, E, H, K, N and Q). Migrating neuroblasts and developing neurons express high levels of the microtubule-associated protein doublecortin (DCX) during the initial phase of morphological maturation (Matsuo et al., 1998; Francis et al., 1999; Gleeson et al., 1999) (DCX; C, F, I, L, O and R). Large species-specific differences in the number and morphology of stained cells are also apparent here. Scale bars: 20 lm. All material has been processed in our laboratory using the same

protocols.


Primates
Cynomolgus monkey (Macaca fascicularis)
Rhesus monkey (Macaca mulatta)
Carnivora
Red fox (Vulpes vulpes)
Lab mouse
C57BL/6J
ICR (CD1)
Lab rat
Fischer 344
Sprague-Dawley
Other rodents (wild and lab)
Long tailed wood mouse (Apodemus sylvaticus)
Cairo spiny mouse (Acomys cahirinus)
Bank vole (Myodes glareolus)
Young neurons per total granlue cells
Age (months)
0
50
100
150
200
250
300
0
1
2
3
4
5


ln (young neurons per total granule cells)
ln (age)
−4
−3
−2
−1
0
1
2
3
4
5
6
−3
−2
−1
0
1
2

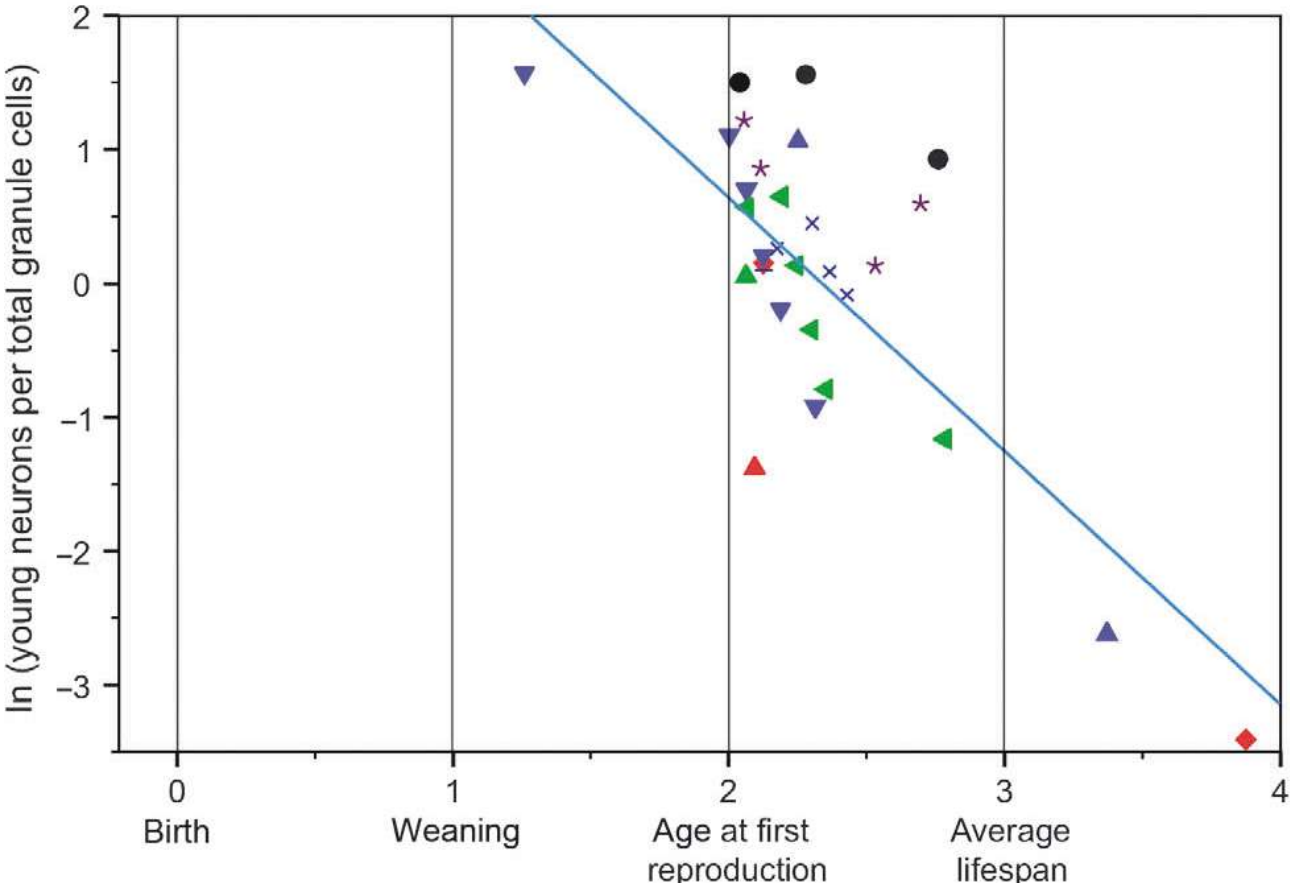


Fig. 2. Hippocampal cell proliferation. (A) Common exponential decline of normalized numbers of proliferating cells (proliferation as a percentage of total granule cell number) during ageing of laboratory and wild rodents, foxes and primates. (B) Log-transformed data visualize that the decline in proliferation in primates and rodent follows a similar trend with absolute age [linear model, N = 61 (excluding neonate guinea pigs and neonate rhesus monkeys due to leverage), overall F ratio 74.2, P < 0.0001; b_{age} =)0.94, p_{age} < 0.0001; b_{order} = 0.26, p_{order} = 0.145; $b_{interaction}$ =)0.005, $p_{interaction}$ = 0.951]. (C) Normalizing age with respect to major life history stages (0, birth; 1, weaning; 2, age at first reproduction; 3, average life expectancy) reveals that primates (and foxes) exhibit a lower cell proliferation at each stage than rodents (N = 63, overall F ratio 89.2, P < 0.0001; b_{age} =)1.62, p_{age} < 0.001; b_{order} =)0.74, p_{order} < 0.0001; $b_{interaction}$ = 0.058, $p_{interaction}$ = 0.627). Red line, primates; blue line, rodents. All data were exclusively taken from publications using randomized systematic sampling procedures investigating the dentate gyrus granule cell and subgranular layer in its full septo-temporal extent. In experimental studies, only data from control specimens were taken.
Proliferating cells are visualized with Ki67 or BrdU. Data were collected from the following sources: Kempermann et al. (1997), Sousa et al. (1997), Gould et al. (1999), Cheng et al. (2002), Drapeau et al. (2003), Keuker et al. (2003), Merrill et al. (2003), Amrein et al. (2004a,b), Heine et al. (2004a,b), Roman et al. (2005), Eadie et al. (2005), Guidi et al. (2005), Rao et al. (2006), Kozorovitskiy et al. (2005), Van der Borght et al. (2005a,b,c), Almgren et al. (2007), Leuner et al. (2007), Barker & Galea (2008), Fitting et al. (2008, 2010), Hauser et al. (2009), Amrein & Slomianka (2010), Amrein et al. (2010), Ben Abdallah et al. (2010), Jabe`s et al. (2010), Taffe et al. (2010), Yu et al. (2010) and Aizawa et al. (2011). For detailed information on the data see Supporting Information.

and young differentiating neurons, except for some of the primate data and the guinea pig from which only 5-bromo-2¢-deoxyuridine (BrdU) data are available. No conversion factors have been applied to account for the injection and survival schedule, as pertinent information such as the bioavailability of BrdU and cell cycle length is not known for all species. Other factors, such as the sensitivity and concentration of different antibodies and tissue processing known to influence BrdU counts in rats (Leuner et al., 2009), could not be accounted for in the primate and guinea pig BrdU numbers. We found a reasonable number of studies presenting data for cell proliferation. Cell numbers for young differentiating neurons [doublecortin-(DCX) or Poly-Sialated Neural Cell Adhesion Molecule (PSA-NCAM)-positive cells] are less frequently reported in the literature. To our knowledge, similar measurements do not exist for humans. The one, otherwise straightforward, stereological assessment of cell proliferation in the human hippocampus could not be included here as it comprises total proliferating cells in the molecular layer, dentate gyrus granule cell layer and polymorphic layer together (Boldrini et al., 2009), which would lead to a clear overestimate of proliferation for the dentate gyrus subgranular zone.

Our dataset comprises animals from three mammalian orders. From the order Rodentia, family Muridae, subfamily Murinae, there are data from seven strains of laboratory mice and rats (genus Mus and genus Rattus, respectively) (Steppan et al., 2005), two wild-derived species of the genus Apodemus, the yellow-necked wood mouse (Apodemus flavicollis) and long-tailed wood mouse (Apodemus sylvaticus) (Michaux et al., 2002), and laboratory-raised Cairo spiny mice (Acomys cahirinus) (Frynta et al., 2010). Further, two wild-derived species of the family Cricetidae, subfamily Arvicolinae: bank voles (Myodes glareolus, formerly called Clethrionomys glareolus) and European pine voles (Microtus subterraneus) (Robovsky et al., 2008) are included. From the suborder Hystricomorpha, we include guinea pigs (Cavia porcellus, family Caviidae) (Dunnum & Salazar-Bravo, 2010). From the order Carnivora, family Canidae, the red fox (Vulpes vulpes) (Sillero-Zubiri, 2009) is included. From the order Primates, one member of the family Cebidae, subfamily Callitrichinae, the common marmoset (Callithrix jacchus), and two members of the family Cercopithecidae, subfamily Cercopithecinae, the cynomolgus or long-tailed macaque (Macaca fascicularis,) and rhesus macaque (Macaca mulatta) are included (Groves, 2001). For each species, values of specimens of the same or a very similar age were averaged. Statistical analyses were conducted on log-transformed data of normalized numbers for proliferating cells (Ki67 or BrdU) and young differentiating neurons (DCX or PSA-NCAM). A general linear model was built with proliferating cells as a percentage of total granule cells or differentiating, young neurons as a percentage of total granule cells as response, and log-transformed age, order and the interaction between age and order as effects. Further, to account for differences in life history pace, age was normalized to the major life history stages as follows: 0–1 reflects infancy (birth to weaning), 1–2 reflects adolescence (weaning to age at first reproduction, which is the age at sexual maturity plus gestation length) and 3 reflects the average life span of a species. Values were compiled from the literature and are listed in detail in the Supporting Information.

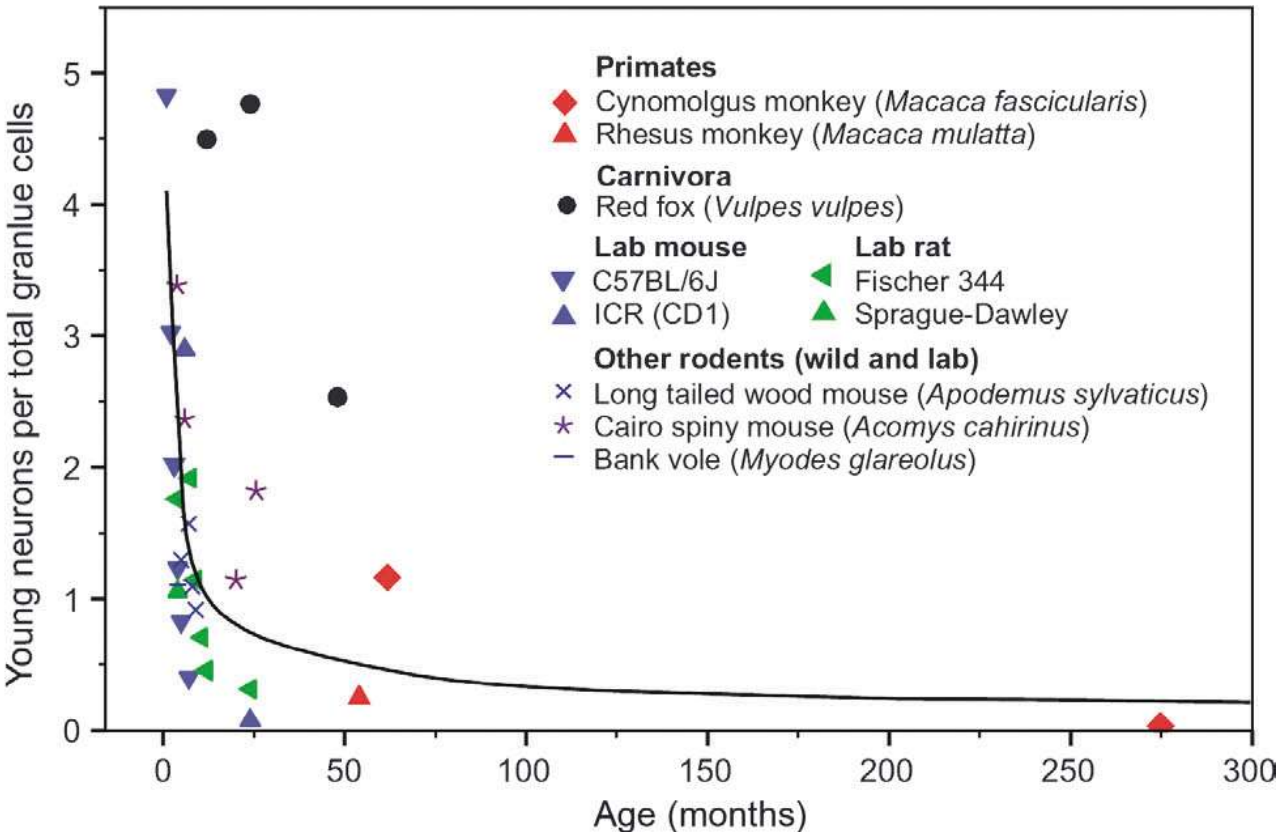
Extent of hippocampal cell proliferation depends strongly on chronological age

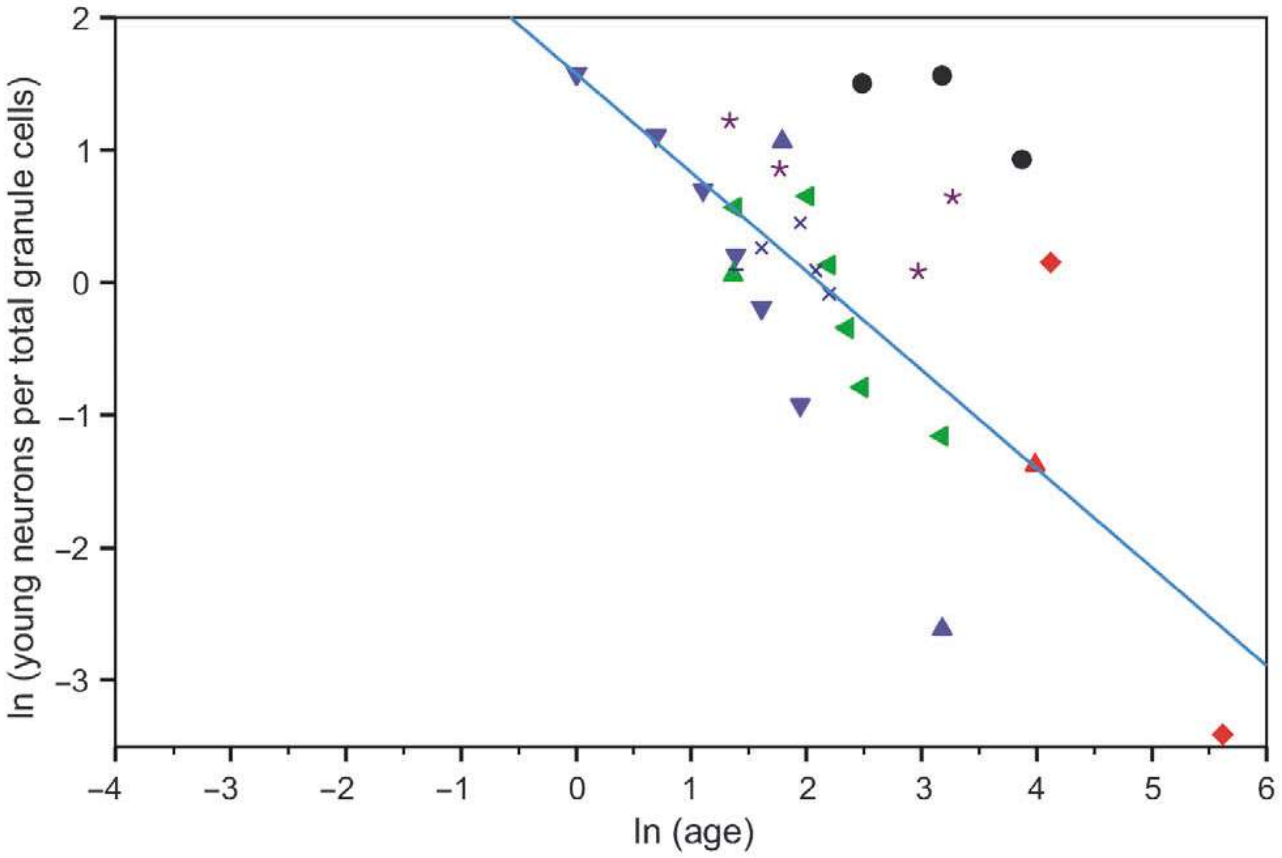
Independent of the order or species, chronological age is a strong predictor for the extent of proliferation in the dentate gyrus. A dramatic exponential decline applies to all investigated species (Fig. 2A). The initially large number of proliferating cells that account for about 5% of the total granule cell population falls below 0.5% within a few months after birth. The plot of logarithmic values reveals some differences between species. Although primates appear to have a higher proliferation rate with respect to absolute age, the difference to rodents is not significant (Fig. 2B). Thus, the ontogenetic differences in cell genesis in early development between primates and rodents do not lead to a significantly altered time course of the decline of postnatal cell genesis. There is no protracted proliferation into adolescence and early adulthood due to long life expectancy. Similarly, the course of the decline in proliferation in precocial species such as guinea pigs and spiny mice is not different from altricial rodents. Normalizing age to the major life history stages of infancy, adolescence and adulthood (Fig. 2C) reveals that primates and foxes show a significantly smaller proliferation rate at any postnatal stage compared with rodents. In summary, the data indicate that, although the life history pace is relatively slow in primates and foxes, cell proliferation declines with absolute age at the same rate as in rodents. Thus, at the time of first reproduction, the number of newly generated cells in the dentate gyrus in long-lived species is downregulated to a great extent. Whether the proliferative potential also decreases is still an open question. For example, ischemia in adult marmosets does not lead to a recruitment of new cells in the dentate gyrus (Bihel et al., 2010), whereas a similar ischemic model in Japanese monkeys does (Tonchev et al., 2003). In contrast, in shortlived mammals, a relatively large number of cells are generated within a period when the animals pass through all of the major ontogenetic events associated with adolescence and adulthood, with their requirements

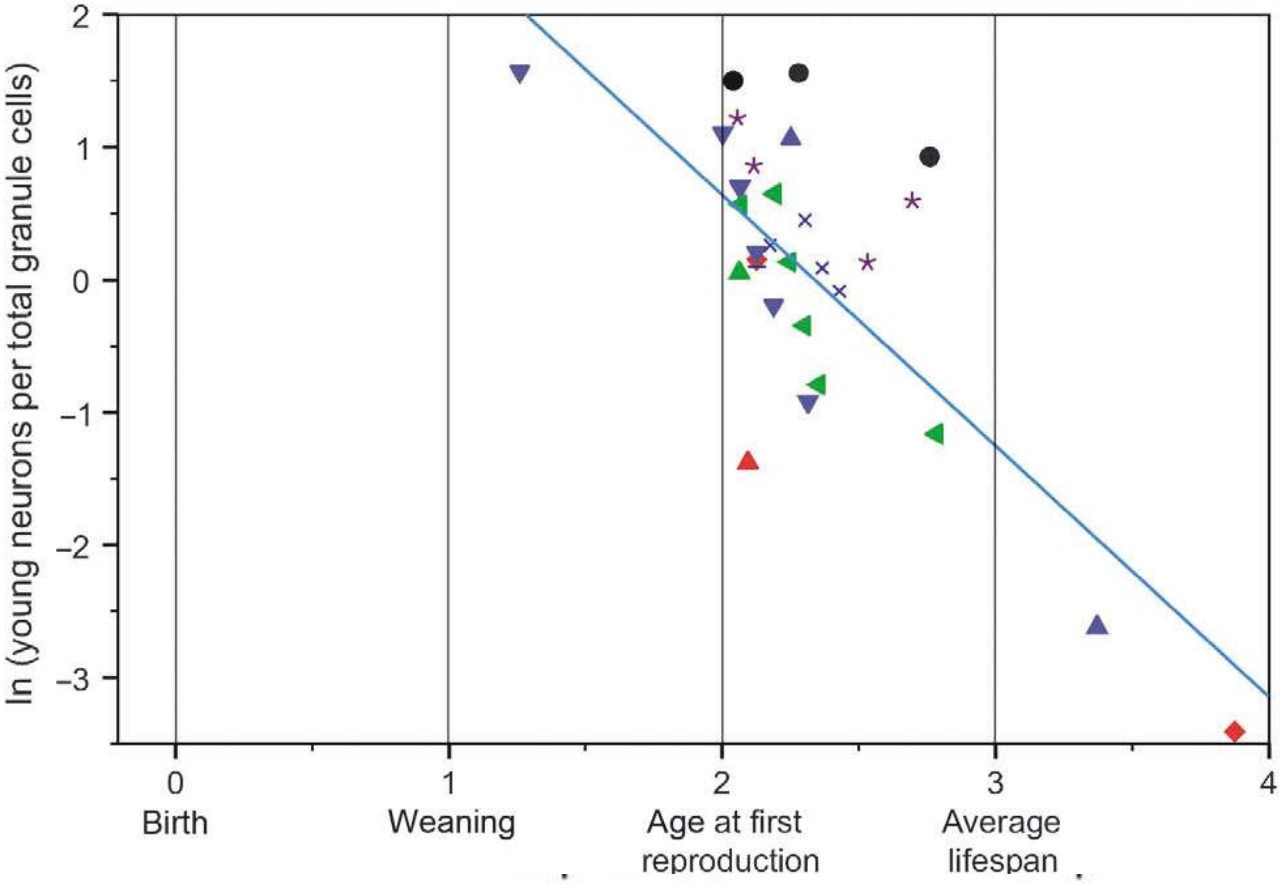
for behavioural flexibility. Rodents can therefore resort to a large pool of newly generated cells to be recruited into the hippocampal circuitry if needed (Ramirez-Amaya et al., 2006; Kee et al., 2007; Snyder et al., 2009; Trouche et al., 2009). In addition, there is a large body of literature showing that rodents possess speciesand strain-specific potential to modulate proliferation at any given age (Abrous et al., 2005). Whether this plasticity in proliferation is typical for laboratory-raised rodents might be an issue for further research. Studies in wild-derived rodents indicate that natural living conditions stabilize proliferation in the face of stimuli that otherwise stimulate proliferation in laboratory rodents, i.e. running and spatial orientation tasks (Hauser et al., 2009; Johnson et al., 2010).

Playground for selective adaptation: neuronal differentiation

In rodents, a common age-dependent decrease of young neurons applies to all species (Fig. 3A and B). The few data points available for primates prohibit a statistical comparison of orders; however, the age-dependent decline of young neurons is also apparent. For normalized age, primates probably exhibit a similar or smaller number of young cells when compared with rodents (Fig. 3B). The same applies to the number of young neurons at life history stages (Fig. 3C). Foxes clearly stand out for the number of young neurons, both for normalized age (Fig. 3B) and life







Age normalized to life history stage

Fig. 3. Young neurons in the dentate gyrus. (A) Normalized numbers of young differentiating neurons [DCX- or PSA-NCAM-positive cells as a percentage of total granule cell number] also decline with age; however, data points show a wider distribution than normalized data for proliferating cells. (B) Log-transformed data show that foxes harbour more cells of the young, maturing stage than rodents or primates (linear model, Rodentia: N = 24, overall F ratio 16.8, $P < 0.001$; age =)0.743, $p_{age} < 0.001$). (C) Normalizing age with respect to major life history stages (0, birth; 1, weaning; 2, age at first reproduction; 3, average life expectancy) reveals that primates exhibit a slightly smaller number of young neurons at each stage than rodents, whereas foxes exhibit a larger number (Rodentia: N = 24, overall F ratio 36.2, $P < 0.0001$; age =)1.91, $p_{age} < 0.0001$). Blue line, rodents. Criteria for including data sets were the same as for Fig. 2. Young neurons are visualized by immunohistochemistry against DCX or PSA-NCAM. Data were collected from the following sources: Kempermann et al. (1997), Cheng et al. (2002), Merrill et al. (2003), Rao et al. (2005, 2006), Fitting et al. (2008, 2010), Barker & Galea (2008), Hauser et al. (2009), Amrein & Slomianka (2010), Amrein et al. (2010), Ben Abdallah et al. (2010), Taffe et al. (2010) and Aizawa et al. (2011). For detailed information on the data see Supporting Information.

history stages (Fig. 3C). Apart from foxes, other species such as spiny mice also show a larger variation in the number of young cells of the neuronal lineage in the dentate gyrus than for proliferation (Fig. 3A). It seems that age is not completely dominating the relation and other factors gain in importance. Regulatory processes that result in variation in the number of young neurons in relation to proliferation could be a decline in cell death, hence increased survival, or an extension of the maturation phase of young neurons as reported in rhesus monkeys (Ngwenya et al., 2006) or aged rats (Rao et al., 2005). Similar ratios of proliferating to dying cells in foxes and rodents also support the extension of the maturation phase in foxes (Amrein&Slomianka, 2010). Increasing the number of functionally distinct, highly excitable young neurons required for specific hippocampal functions (Cameron & McKay, 2001; Deng et al., 2009) by extending the period during which they show these properties appears to be an efficient response to decreased proliferation. Given the data presented here, one might speculate that social and habitat complexity could be parameters increasingly determining neuronal differentiation. In highly social foxes that also show particular behavioural flexibility to environmental conditions, we observe the largest numbers of differentiating neurons in adults. It would be interesting to see if common marmosets as cooperative breeders show a similar increase in number of young granule cells in relation to proliferation, which is, compared with the macaques, relatively low (Fig. 2B and C). In rodents, several types of natural behavioural patterns also influence or correlate with neurogenesis: experiencing maternal care (Bredy et al., 2003), reproductive experience from sex (Leuner et al., 2010) to motherhood (Pawluski & Galea, 2007; Ruscio et al., 2008), aggressive behaviours (Fiore et al., 2005; Veenema et al., 2005) or social interactions (Fowler et al., 2002; Hoshaw et al., 2006; for an in-depth review see Gheusi et al., 2009). Interestingly, the highly social, food-sharing spiny mice (Porter et al., 1981; Novakova et al., 2008) also maintain large numbers of young neurons during ageing (Fig. 3B and C) (Amrein et al., 2010).

Implications for short- and long-lived mammals

Small rodent species usually have a short life span. Laboratory mice can live up to 2 years, and rats have an average life span of 2.5 years (see Supporting Information). In the wild, only a fraction of small rodents survive into the second year of life (Berry et al., 1973), although record longevities of several years in captivity have been observed. As a consequence, rodents reach sexual maturity early in life. The time window of maximal behavioural flexibility that is characteristic for adolescence therefore overlaps with the time of first breeding. This is also the time window when cell proliferation as well as the number of young neurons is still at a relatively high rate (Figs 2C and 3C). In primates, the age at first reproduction varies from 1 year in small lemurs to more than 15 years in orangutans, and the primates for which adult neurogenesis has been investigated first reproduce between the ages of 20 months (common marmosets) and 42–46 months (rhesus and cynomolgus monkeys, respectively, although wild populations may show even higher values) (Jiang et al., 1989; Ross, 1991; van Noordwijk & van Schaik, 1999; van Schaik & Isler, in press). At this

age, the down-regulation of proliferation has proceeded to leave only a fraction of the newly formed cells that were observed around weaning (Fig. 2C). Foxes reach sexual maturity at a minimum age of 10 months (Sillero-Zubiri, 2009; Nelson & Chapman, 1982), but as data for juveniles are lacking, we can only speculate that proliferation rates would also be higher before adulthood.

The surprisingly similar time course of the exponential decline in proliferation with absolute age within species and between orders indicates that this process is independent of life history pace and the timing of specific ontogenetic periods of a species. Nevertheless, individual species can differ from each other considerably at any given time point. Thus, functional effects attributed to adult neurogenesis may well differ between species as they reach ontogenetically important events at different points along their chronological axis of life. Distinct species differences on the background of a strong age effect resemble the Finlay–Oxnard debate on brain size. Total brain size explains the size of individual components of the brain better than ecology or evolution (Finlay & Darlington, 1995). However, the size of brain components relative to each other can define both phylogenetic groups and groups showing similar ecological adaptations (de Winter & Oxnard, 2001). Similar to the major effects of total brain size on the size of its components, the effect of age appears to have an overwhelming effect on cell proliferation, but less so on neuronal differentiation. The fine-tuning of neuronal differentiation, be it by increasing survival and/or the duration of maturation of the young neurons, might be the adaptive answer to species-specific needs, and could therefore be the field where we might find important insights to the still-debated question of the functional relevance of adult neurogenesis in the dentate gyrus.

Supporting Information

Additional supporting information can be found in the online version of this article:

Table S1. Data compilation of total proliferating cells, immature neurons and granule cell numbers.

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Abbreviations

BrdU, 5-bromo-2'-deoxyuridine; DCX, doublecortin; PSA-NCAM, Poly-Sialated Neural Cell Adhesion Molecule.

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